

## CLAIMS

What is claimed is:

1. A chimeric antibody, comprising at least part of a human immunoglobulin constant region and at least part of a non-human immunoglobulin variable region, said antibody capable of binding an epitope specific for human tumor necrosis factor TNF  $\alpha$ .  
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2. A chimeric antibody according to claim 1, wherein said binding of said antibody to TNF $\alpha$  inhibits a pathologic activity of TNF $\alpha$ .
3. A chimeric antibody according to claim 1, wherein said antibody does not bind to one or more epitopes included in amino acids 11-13, 37-42, 49-57 or 155-157  
10 of SEQ ID NO: 1.
4. A chimeric antibody according to claim 1, wherein said chimeric antibody comprises two light chains and two heavy chains, each of said chains comprising at least part of a constant region and at least part of a variable region, said  
15 variable region capable of binding an epitope of human TNF $\alpha$ .
5. A chimeric antibody according to claim 1, wherein said antibody neutralizes human TNF $\alpha$  under physiological conditions.
6. A chimeric antibody according to claim 1, wherein said variable region is of murine origin.

7. A chimeric antibody according to claim 1, wherein said variable region is derived from a high affinity murine monoclonal antibody which binds to a neutralizing epitope of human TNF $\alpha$ .
8. A chimeric antibody according to claim 7, wherein said murine monoclonal antibody competitively inhibits the binding of monoclonal antibody cA2 to TNF $\alpha$ .  
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9. A chimeric antibody according to claim 1, characterized by an affinity, measured as an association constant (Ka), of at least  $1 \times 10^8$  liter/mole.
10. A chimeric antibody according to claim 9, wherein said affinity, measured as an association constant (Ka), is at least  $1 \times 10^9$  liter/mole.  
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11. A chimeric antibody according to claim 1, wherein said antibody neutralizes human TNF $\alpha$  with an ID50 of at least 1  $\mu$ g/ml.
12. A chimeric antibody according to claim 11, wherein said antibody neutralizes human TNF $\alpha$  with an ID50 of at least 15 ng/ml.  
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13. A chimeric antibody according to claim 12, wherein said antibody neutralizes human TNF $\alpha$  with an ID50 of at least about 100 ng/ml.
14. A chimeric antibody according to claim 1, wherein said antibody is in detectably labeled form.
15. A chimeric antibody according to claim 1, wherein said antibody is produced recombinantly.  
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16. An immunoassay method for detecting human TNF in a sample, comprising:
  - (a) contacting said sample with an antibody according to claim 36, or a TNF binding fragment thereof, in detectably labeled form; and
  - (b) detecting the binding of the antibody to said TNF.
- 5 17. The chimeric antibody cA2.
18. A chimeric antibody, comprising at least part of a human IgG1 constant region and at least part of a non-human immunoglobulin variable region, said antibody capable of binding an epitope specific for human TNF $\alpha$ .
- 10 19. A chimeric antibody according to claim 18, wherein said binding of said antibody to TNF $\alpha$  inhibits a pathologic activity of TNF $\alpha$ .
20. A chimeric antibody according to claim 18, wherein said antibody does not bind to one or more epitopes included in amino acids 11-13, 37-42, 49-57 or 155-157 of SEQ ID NO: 1.
- 15 21. A chimeric antibody according to claim 18, wherein said chimeric antibody comprises two light chains and two heavy chains, each of said chains comprising at least part of a constant region and at least part of a variable region, said variable region capable of binding an epitope of human TNF $\alpha$ .
22. A chimeric antibody according to claim 18, wherein said antibody neutralizes human TNF $\alpha$  under physiological conditions.
- 20 23. A chimeric antibody according to claim 18, wherein said variable region is of murine origin.

24. A chimeric antibody according to claim 18, wherein said variable region is derived from a high affinity murine monoclonal antibody which binds to a neutralizing epitope of human TNF $\alpha$ .
- 5        25. A chimeric antibody according to claim 24, wherein said murine monoclonal antibody competitively inhibits the binding of monoclonal antibody cA2 to TNF $\alpha$ .
- 10      26. A chimeric antibody according to claim 18, characterized by an affinity, measured as an association constant (Ka), of at least  $1 \times 10^8$  liter/mole.
- 15      27. A chimeric antibody according to claim 26, wherein said affinity, measured as an association constant (Ka), is at least  $1 \times 10^9$  liter/mole.
- 20      28. A chimeric antibody according to claim 18, wherein said antibody neutralizes human TNF $\alpha$  with an ID50 of at least 1  $\mu\text{g}/\text{ml}$ .
29. A chimeric antibody according to claim 28, wherein said antibody neutralizes human TNF $\alpha$  with an ID50 of at least 15 ng/ml.
- 15      30. A chimeric antibody according to claim 28, wherein said antibody neutralizes human TNF $\alpha$  with an ID50 of at least about 100 ng/ml.
31. A chimeric antibody according to claim 18, wherein said antibody is in detectably labeled form.
- 20      32. A chimeric antibody according to claim 18, wherein said antibody is produced recombinantly.

33. An immunoassay method for detecting human TNF in a sample, comprising:

- contacting said sample with an antibody according to claim 38, or a TNF binding fragment thereof, in detectably labeled form; and
- detecting the binding of the antibody to said TNF.

5 34. A chimeric antibody, comprising two light chains and two heavy chains, each of said chains comprising at least part of a human immunoglobulin constant region and at least part of a non-human immunoglobulin variable region, said variable region capable of binding an epitope of human tumor necrosis factor hTNF $\alpha$ , wherein said light chains comprise variable regions comprising SEQ ID NO: 3 and said heavy chains comprise variable regions comprising SEQ ID NO: 5.

10 35. A chimeric antibody according to claim 34, wherein the human immunoglobulin constant region is an IgG1

15 36. A chimeric antibody comprising at least part of a human immunoglobulin constant region and at least part of a non-human immunoglobulin variable region, said antibody capable of binding an epitope specific for human tumor necrosis factor TNF $\alpha$ , wherein the non-human immunoglobulin variable region comprises an amino acid sequence selected from the group consisting of SEQ ID NO: 3 and SEQ ID NO: 5.

20 37. A chimeric antibody comprising at least part of a human IgG1 constant region and at least part of a non-human immunoglobulin variable region, said antibody capable of binding an epitope specific for human TNF $\alpha$ , wherein the non-human immunoglobulin variable region comprises an amino acid sequence selected from the group consisting of SEQ ID NO: 3 and SEQ ID NO: 5.

38. A chimeric antibody comprising at least part of a human immunoglobulin constant region and at least part of a non-human immunoglobulin variable region, said antibody capable of binding an epitope specific for human tumor necrosis factor TNF $\alpha$ , wherein the non-human immunoglobulin variable region comprises a polypeptide encoded by a nucleic acid sequence selected from the group consisting of SEQ ID NO: 2 and SEQ ID NO: 4.

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39. A chimeric antibody comprising at least part of a human IgG1 constant region and at least part of a non-human immunoglobulin variable region, said antibody capable of binding an epitope specific for human TNF $\alpha$ , wherein the non-human immunoglobulin variable region comprises a polypeptide encoded by a nucleic acid sequence selected from the group consisting of SEQ ID NO: 2 and SEQ ID NO: 4.

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40. A polypeptide comprising the amino acid sequence selected from the group consisting of SEQ ID NO: 3 and SEQ ID NO: 5 or at least one binding fragment thereof, wherein said polypeptide binds to hTNF $\alpha$ .

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41. A polypeptide of Claim 40, wherein said polypeptide neutralizes hTNF $\alpha$ .

42. A polypeptide of Claim 40, wherein said polypeptide inhibits hTNF $\alpha$ .

43. A polypeptide of Claim 40 which binds to at least one epitope included in amino acids 87-108, or both 59-80 and 87-108, of SEQ ID NO: 1.

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44. A polypeptide of Claim 40 which does not bind to an epitope included in amino acids 11-13, 37-42, 49-57 or 155-157 of SEQ ID NO: 1.

45. A polypeptide of Claim 40 which competitively inhibits the binding of monoclonal antibody cA2 to hTNF $\alpha$ .

46. A polypeptide comprising the amino acid sequence of SEQ ID NO: 3, wherein said polypeptide binds to hTNF $\alpha$  and competitively inhibits the binding of monoclonal antibody cA2 to hTNF $\alpha$ .

5 47. A polypeptide comprising the amino acid sequence of SEQ ID NO: 5, wherein said polypeptide binds to hTNF $\alpha$  and competitively inhibits the binding of monoclonal antibody cA2 to hTNF $\alpha$ .

10 48. A polypeptide comprising at least one binding fragment of SEQ ID NO: 3, wherein said polypeptide binds to hTNF $\alpha$  and competitively inhibits the binding of monoclonal antibody cA2 to hTNF $\alpha$ .

49. A polypeptide comprising at least one binding fragment of SEQ ID NO: 5, wherein said polypeptide binds to hTNF $\alpha$  and competitively inhibits the binding of monoclonal antibody cA2 to hTNF $\alpha$ .

15 50. A polypeptide of Claim 45 having a hTNF $\alpha$  binding affinity, measured as an affinity constant (Ka), of at least  $1 \times 10^8$  liters/mole.

51. A polypeptide of Claim 45 having a hTNF $\alpha$  binding affinity, measured as an affinity constant (Ka), of at least  $1 \times 10^9$  liters/mole.

20 52. A polypeptide of Claim 45 which neutralizes hTNF $\alpha$  with an ID50 of at least about 1  $\mu$ g/ml.

53. A polypeptide of Claim 45 which neutralizes hTNF $\alpha$  with an ID50 of at least about 100 ng/ml.
54. A polypeptide of Claim 45 which neutralizes hTNF $\alpha$  with an ID50 of at least about 15 ng/ml.
- 5 55. A fusion protein comprising SEQ ID NO: 3 or SEQ ID NO: 5 or a binding fragment thereof, wherein said fusion protein binds to hTNF $\alpha$ .